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on February 13, 2004

  
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**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

Applicant: Bodor et al.  
Serial No.: 10/046,470  
Filed: November 20, 2001  
For: FOOD PRODUCT COMPRISING CAROTENOIDS

Edgewater, New Jersey 07020  
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**SUBMISSION OF PRIORITY DOCUMENT**

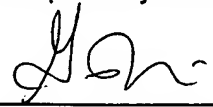
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Sir:

Pursuant to rule 55(b) of the Rules of Practice in Patent Cases, Applicant(s) is/are submitting herewith a certified copy of the European Application No. 00310404.9 filed November 24, 2000, upon which the claim for priority under 35 U.S.C. § 119 was made in the United States.

It is respectfully requested that the priority document be made part of the file history.

Respectfully submitted,

  
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Die angehefteten Unterlagen stimmen mit der ursprünglich eingereichten Fassung der auf dem nächsten Blatt bezeichneten europäischen Patentanmeldung überein.

The attached documents are exact copies of the European patent application described on the following page, as originally filed.

Les documents fixés à cette attestation sont conformes à la version initialement déposée de la demande de brevet européen spécifiée à la page suivante.

**Patentanmeldung Nr.    Patent application No.    Demande de brevet n°**

00310404.9

Der Präsident des Europäischen Patentamts;  
Im Auftrag

For the President of the European Patent Office

Le Président de l'Office européen des brevets  
p.o.

**I.L.C. HATTEN-HECKMAN**

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**Blatt 2 der Bescheinigung  
Sheet 2 of the certificate  
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**FOOD PRODUCT COMPRISING CAROTENOIDS****Field of the invention**

5 The present invention relates to an edible composition, preferably an edible emulsion, comprising a colored compound, especially carotenoids and/or derivatives thereof.

**10 Background of the invention**

Carotenoids, in particular, beta carotene, are well known and have been reported to protect cells from the reactive species (free radicals) that are suggested to play a role  
15 in immunity and cell-cell communication. It has also been reported to be an anti-ageing agent in human cells, and so slowing down the ageing process of the cells of humans. Supplementation of food products with one of the most commonly known carotenoids, beta carotene, is well known.  
20 Beta carotene is a well known source as it is the pro-vitamin of vitamin A. It is furthermore used as a colorant in food products, for example to colour drinks, soups, tomato products and margarine type of products. In such products, as a general indication, amounts around 5  
25 mg/kg are used. Such an amount changes the colour of a white product to be light yellow. It will be appreciated that regular amounts can vary among different food product. Food products comprising around 5 mg/kg of carotenoids and no or few other colored compounds  
30 are generally pale yellow of colour. The higher the carotenoid content, the more intense the colour is.

In particular in the Western world, consumers are reluctant to products of which the colour has changed from what they are used to. For example, margarine, butter, halvarin or salad oils , as well as mayonnaise, are considered  
5 unacceptable by consumers when the colour of such products is intense yellow, orange or even red. However, at the same time, this type of products have been found by applicants to be excellent vehicles of the daily intake of amounts of carotenoids sufficient to obtain an elevated carotenoid  
10 status in human blood serum levels.

Hence, a desire to add carotenoids to food products in amounts higher than needed for colouring, or in amounts above which any change in colour of the food product is  
15 still desirable or acceptable, is present.

In particular, there is a desire for food products that are fortified with colored compounds like carotenoids and/or derivatives thereof, while the colour is comparable to the  
20 colour of regular products of the same composition, that are not fortified with these colored compounds.

Fortification is nowadays also desirable for food products which may lead to a reduction of the body carotenoids by their consumption, for example in food products which  
25 contain sucrose polyesters which are used as fat replacers, or contain materials like sterols and stanols, or esters thereof.

Fortification of food products with carotenoids or  
30 derivatives is known. For example EP-A-671,461 discloses the addition of significant amounts of carotenoids to oil.



In WO-A-95/05747 the use of beta carotene to reduce oxidation of fats and oils is disclosed. Compositions fortified with carotenoids are furthermore disclosed in EP-A-354,600.

5

Several attempts were made in the past to apply enhanced amounts of carotenoids to food products without significantly changing the colour of the food products.

- 10 US 5,532,009 discloses food products fortified with a water soluble carotenoid/cyclodextrin complex. It is disclosed that the carotenoid is encapsulated in cyclodextrin. Although it is reported that colour intensity and degradation are minimized, we have found that dextrin  
15 encapsulation can not sufficiently serve to reduce the yellow and/or red colour intensity of products comprising increased levels of carotenoids.

#### Field of the invention

- 20 Compounds belonging to the class of carotenoids are best known for their role as dietary antioxidants, although other potentially protective mechanisms for this group of compounds have been identified e.g. provitamin A activity. About 600 carotenoids occur in nature of which the six  
25 major ones in the diet are  $\beta$ -carotene, lycopene, lutein,  $\beta$ -cryptoxanthin,  $\alpha$ -carotene and zeaxanthin. Although no thorough scientific base for claiming a direct relation between cause and effect of the carotenoid intake and disease incidence has been proven thus far, inverse  
30 associations between carotenoid intake or status and disease incidence have been found.

We have now found edible compositions which can contain a significant amount of carotenoids, i.e. an amount of carotenoids substantially higher than found these days in such products, but without a significant change of colour due to this increased amount of carotenoids, of these edible compositions. Preferably, the compositions are emulsions.

Hence, emulsions according to the present invention contain a carotenoids level of at least 15 mg/kg, and preferably at least 18 mg/kg, and have a yellowness factor as defined in this specification, of less than 4000, and have a yellowness index in the range of 1 to 90. Though higher levels than 150 mg/kg of carotenoids can be applied, it has been found that there is hardly any contribution to any of the benefits indicated of these amounts over such a level. Of course, highest amounts applicable depend on the amount of food taken on a daily basis. For example, a single portion of a margarine varies in different countries. But differences in daily portions is even larger between the different food products the carotenoids are applicable in. The carotenoids are evenly distributed throughout the composition. In case the carotenoids are present in a food product or emulsion in encapsulated form, evenly distributed means that the encapsulates are evenly distributed throughout the product or emulsion. This means that, if part of the composition is consumed, the level of carotenoids in the part (whichever one this is, and whichever the size of the part) will be as indicated. The carotenoids being evenly distributed also means that food products, in which in only a part thereof, a significantly increased concentration of carotenoids, compared to other

parts of the same food product, is found, is not incorporated in this invention. An advantage with the compositions of this invention is that if only a small portion of the edible composition is consumed, still an  
5 increased level of carotenoids is eaten. Hence, in all embodiments of this invention, the carotenoids are homogeneously distributed throughout the composition. In case encapsulates are applied according to this invention, the distribution of the encapsulates is such that the  
10 number of encapsulates is roughly the same throughout each part of the edible composition.

In a further preferred embodiment, products according to the present invention contain a carotenoids level of at  
15 least 15 mg/kg, and preferably at least 18 mg/kg, and have a yellowness factor as defined in this specification, of less than 4000, and the yellowness index is in the range of 1 to 75.

20 The yellowness factor is the yellowness index, divided by the carotenoids level expressed in g/kg.  
The yellowness index is determined from samples at 15 °C, on a Tricolor LFM3 Colorimeter (Dr Lange). The Yellowness Index is calculated from the tristimulus colour (in X, Y  
25 and Z) of the sample, referred to the standard illuminant C (white plate) and the 2 °C observer according to the formula

$$Y.I. = \frac{1.28 \times X - 1.06 \times Z}{Y} \times 100$$

30

Such food products can show several benefits:

- the presence of an elevated amount of one or more compounds which have a strong coloring capacity, which compounds provide health benefits
- 5 • a colour comparable to that of similar products which do not contain such an elevated amount of one or more of these healthy compounds.

10 An edible composition has now been found that comprises increased levels of carotenoids.

#### Summary of the invention

15 Preferably, compositions of the invention contain at least 15 mg/kg carotenoids, more preferred at least 18 mg/kg carotenoids, and have a yellowness factor of less than 3333, more preferred of less than 2850, and a yellowness index which is in the range of 1 to 65.

20 In particular, preferred embodiments within the current invention compositions contain at least 20 mg/kg, and have a yellowness factor of less than 2000, the yellowness index being in the range of 1 to 70.

25 Particular embodiments in which the edible composition or food products contain more than 35 mg/kg carotenoids, and have a yellowness factor of less than 2000 and a yellowness index in the range of 1 to 70 can fulfill specific needs for food products which contain significant amounts of carotenoids, without these food products having altered  
30 significantly compared to similar products not containing these high amounts of carotenoids. This is even more the

case for products containing even more than 45 mg/kg with a yellowness factor of less than 2000 and a yellowness index of less than 90.

- 5 In a second aspect the invention relates to particular food products fulfilling these criteria, methods for preparing such food products, and carotenoid containing compositions for use in products so as to obtain products according to the specifications indicated above.

10

#### Detailed description of the invention

Where weight percentages are indicated throughout this application, they are expressed as wt% on total product weight, unless indicated otherwise.

15

Edible compositions according to the invention comprise colored compounds, said colored compounds being carotenoids and/or derivatives thereof. Derivatives are for example chemically modified carotenoids that still show the desired anti-oxidant functionality.

20

In this patent specification, where reference is made to carotenoids, both carotenoids and derivatives thereof are meant.

25

Preferred carotenoids are  $\beta$ - carotene, lycopene, lutein,  $\beta$ -cryptoxanthin,  $\alpha$ -carotene and zeaxanthin.

The most preferred carotenoids are  $\beta$ - carotene,  $\alpha$ -carotene, lutein and lycopene.

30

For all embodiments within the present invention, it is preferred that the edible composition is an edible emulsions, as with food emulsions, the problem of increasing color upon increasing carotenoids level, and the undesirability thereof is largest. In many cases this type of product is in most cases desired as a white to pale yellowish product.

In a particular embodiment, the compositions contain a carotenoid mixture of lutein, lycopene, and  $\beta$ - and  $\alpha$ -carotene. The use of palm oil carotene is preferred over synthetically obtained carotene. In a further preferred embodiment, the edible composition contains between 15-40 mg/kg carotenoids, of which about  $2/3^{\text{rd}}$  of the carotenoid is lutein, about  $1/6^{\text{th}}$  is lycopene, with the remaining part being a mixture of  $\beta$ - and  $\alpha$ -carotene. The  $\beta$ - and  $\alpha$ -carotene mixture in this preferred embodiment is preferably obtained from a natural source, of which palm-oil is a suitable example.

Said colored carotenoids compounds and/or derivatives thereof can be used in free form, or can be present in capsules, hence being present in encapsulated form.

In yet another embodiment, also free colored compounds like carotenoids can be present in addition to the encapsulated carotenoids. However it is preferred to keep the free colored compound content as low as possible.

Several methods have been found to obtain the edible compositions of the current invention and food products consisting of, or containing such edible compositions.

In a particular embodiment of the invention, carotenoids are encapsulated. This embodiment is preferred over other embodiments.

Encapsulated form means said colored compounds like  
5 carotenoids and/or derivatives thereof are at least partly, preferably fully retained in a capsule. The term encapsulated both refers to an embodiment wherein a thin coating (capsule wall) is formed around the encapsulates (for example carotenoids), and to an embodiment wherein the  
10 encapsulates are trapped within or throughout a matrix.

The term "encapsulates" is also used to indicate encapsulated carotenoids.

Several methods for encapsulating carotenoids have been  
15 found which can be used for the preparation of the compositions according to the invention. Suitable encapsulation methods include encapsulation with a protein layer, encapsulation through the preparation of complex coacervates, preparation of duplex emulsions, and the like.  
20 It has been found that with these encapsulation methods, a further improvement can be obtained by the addition of minor amounts of titanium dioxide in the coating or matrix material of the particle. The amount of titanium dioxide per  
25 particle depends on the size of the capsule and the amount of carotenoid which is to be enclosed in it. Suitably, 5-30 wt% of titanium dioxide, and preferably 5-20 wt% of titanium dioxide, based on the total weight of the particle is applied.

30 The encapsulated form can for example be embodied by encapsulation of the colored compounds like carotenoids

10

and/or derivatives thereof in crosslinked compounds, precipitated compounds or heat set compounds.

The products of the invention can be obtained by the addition of relatively large particles. Such particles are preferably, but not necessarily smaller than 300  $\mu\text{m}$ . (average size of the particles present), and preferably are less than 150  $\mu\text{m}$ , more preferably less than 50  $\mu\text{m}$ .

The size of the capsules would depend on the desired effect. However, to obtain products which appear not to contain a carotenoid fortification, i.e. which contain increased amounts of carotenoids, without the consumer using the product clearly noticing a difference, suitably contain capsules which have a particle size of less than 50  $\mu\text{m}$ , preferably less than 30  $\mu\text{m}$ , most preferred from 1 to 20  $\mu\text{m}$ .

Preferably encapsulation of the colored compounds like carotenoids and/or derivatives thereof is in compounds which are degradable in the human body.

Carotenoids for use in edible compositions of the current invention can, for example, be encapsulated in proteins. Suitable encapsulation can for example be obtained by crosslinked proteins on a fat globule surface.

Optionally said crosslinking is obtained under application of an enzyme suitable as a catalyst for crosslinking of said protein or other compounds. Suitable enzymes are for example transglutaminases, peroxidases, laccases, tyrosinases or combinations thereof. The selection of



enzyme is believed to be related to the protein or compound that is used as a substrate for crosslinking as some of the mentioned enzymes are known to be substrate specific. A preferred combination is transglutaminase with protein,  
5 with the proviso that whey protein if used is preferably in the denatured state.

In a preferred embodiment colored compounds like carotenoids and/or derivatives thereof are encapsulated in  
10 a shell consisting essentially of heat set protein.

Preferably said encapsulates are protein coated fat globules, whereby carotenoids are dissolved in said fat. The thus formed encapsulates can be made visible by  
15 microscopy and protein staining.

The presence of protein coated fat globules can also be demonstrated by a method wherein the edible composition comprising encapsulates is heated (for example to about 50 °C) and centrifuged. The protein content in the bottom  
20 layer is then determined. This is protein in solution. The protein in the creamed fat layer can also be determined. It will be appreciated that protein capsules or shells obtained by way of a combination of any of the above methods such as heating in combination with  
25 transglutaminase treatment are also encompassed. It has been found that even stronger and more stable particles are obtained if more than one layer of protein is applied as coating of the carotene encapsulates.

30 Another suitable way to obtain encapsulation can be by encapsulation in edible polymers other than protein,

whereby dextrans are excluded. Examples of such polymers are gelatin, starch and calcium alginate polymers.

Encapsulation can be in the form of coacervates.

For encapsulation, in most embodiments it is preferred to  
5 dissolve and/or disperse the carotenoid or carotenoid mixture in an oil or wax, which oil or wax is then added to an aqueous solution of polymer or polymer mixture (for complex coacervates) and treated so as to obtain the encapsulate. Setting of the polymer mixture so as to obtain  
10 particles can be provided for by reduction of temperature, by crosslinking by the use of an enzyme or chemical compound, or, for example, by adjusting the pH, e.g., to levels below 6, preferably to a level in the range between 4 and 5. The most suitable way will depend on the polymers  
15 used.

In view of their resistance to high shear conditions, storage stability when applied in an emulsion, and capability of reduction of colour, preference is found for particles of complex coacervates. For the preparation of  
20 such complex coacervates, reference is made to EP 790780, though for the current invention, also carotenoids are present as indicated above.

It has been found that the use of such particles, for example of gelatin and Arabic gum, crosslinked by use of  
25 glutaraldehyde, provide very good, stable particles. By the addition of titaniumdioxide in the coacervation process the titaniumdioxide is included in the particles. These particles of carotenoids in complex coacervates also including 5-30 wt%, preferably 5-20 wt%, based on the  
30 particle weight, of titaniumdioxide, are preferred. Such particles are capable of obtaining a significant reduction

of carotenoids colour in food products, preferably edible emulsions, when compared to non-encapsulated carotenoids in such products. The particles so obtained can be applied at levels of carotenoids up to 40 times as much as present with products known in the art without a significant change of colour, compared to products comprising these amounts of carotenoids without any treatment.

In yet another embodiment of the invention, part or all of the carotenoids are present in free form, and the composition in which the carotenoids are applied are aerated.

Colored compounds like carotenoids in free form are colored compounds that are not encapsulated or at least present in such a form that the colour of these compositions is clearly visible in a product comprising these colored compounds.

The reduction of colour intensity for such free carotenoids containing products is suitably achieved by aerating these products, for example by sparging the products, after their preparation, with a suitable, non-toxic gas. Examples of such gases include air, oxygen, and, preferably, nitrogen. Aeration should be at least 10%. It has been found that with aeration of 30 up to 50% products can be obtained which are widely acceptable by the consumers. Such products differ in carotenoid level and aeration level, but are yet perceived as being products similar in colour to the products these consumers are used to. Aeration is a method in particular suitable for fat based food products of a consistency which allows maintenance of the air

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bubbles in the product. Good examples of such products include spreads such as butter, margarines and lower fat spreads, or mixtures thereof, cream cheeses or other soft cheeses, ice cream, and particular kinds of dressings and sauces, desserts, mousses, and the like.

Alternatively, very small water droplets can be added to the emulsion for colour reduction in a manner similar to that of aeration. This method is in particular suitable for water in oil emulsions. The very small water droplets preferably do not contain any further coloring ingredients.

In yet another embodiment, an edible composition is prepared in which part of the carotenoids are present in encapsulated form and part are present in free form. In this embodiment, the level of free carotenoids can be adjusted so as to set the desired colour of the food product. For example, a product can be prepared in which lutein encapsulates, lycopene encapsulates, and free carotene are used. In a particular embodiment, such a food product can contain 5-15 mg/kg of lutein , 1-8 mg/kg of lycopene, present as separate encapsulates or as encapsulates containing both, and 3-8 mg/kg of a mixture of alpha and beta carotene in free form.

Also encompassed within the current invention are encapsulates containing carotenoids in which the carotenoids are encapsulated in complex coacervates of at least two polymers, and the encapsulates contain 5-30 wt% on particle weight, of titaniumdioxide.

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It is furthermore desired that the free carotenoids and/or encapsulates are evenly distributed throughout the edible composition. Such can suitably be obtained by adequate mixing. Though homogenization can be applied, for some encapsulates, such as protein encapsulates, this method is less preferred as high shear pressures could lead to breaking of the particles and hence coloring of the product if done at high pressures.

10 Edible compositions according to the invention are characterised by a maximum Yellowness Index whilst containing a significant amount of carotenoids. When applying these high amounts of carotenoid to food products of the same composition, but without any technical measure, 15 the colour of such 'untreated' food products will be significantly higher than the yellowness index of the products of the current invention. Without any treatment the Yellowness Index will be at least 10% higher. In a specific and preferred embodiment, the edible compositions are used for the preparation of food products, 20 and preferably of edible emulsions, and more preferred these are ready-to-eat food products. Food products encompassed in the invention are preferred to be common products which are often used by consumers on a 25 daily basis in amounts different for each individual. The invention is especially applicable for fat based food products. Fat based food products are food products (partially) based on fat and regarded by the consumer as "fatty type of products".

30

Such fat based food products can be for example water free food products such as shortenings or cooking fats. Said fat based food products can also be water containing food products, which can be for example water continuous or fat continuous or bicontinuous. Examples of water continuous products are cheese, dressings, including mayonnaise, ice cream, milk type drinks, (drink) yoghurt, toppings and fillings, low fat margarine like 40% or 20% fat spreads. Examples of fat continuous products are margarine and butter, low fat margarines containing for example 40 or 25% fat, and the like. Suitable fat based products are for example liquid margarines, which are pourable or pumpable products comprising generally from 60 to 95 wt% fat. Bicontinuous oil and water containing products are for example disclosed in EP-A-463,688.

These preferred products according to the invention can be of any consistency, for example pourable such as coffee creamer, pumpable such as certain liquid margarines, squeezable such as food pastes, spreadable such as margarine or margarine like table spreads, spoonable such as dairy creams, or wrapper type of products such as shortenings.

Fat based food products of high carotenoids contained without change of colour are preferably chosen from the group of shortenings, cooking fats, cheese, dressings, including mayonnaise, ice cream, milk type drinks, (drink) yoghurt, toppings and fillings, butter, margarine and low fat margarine. Products thereof used in the western world on a daily basis and so very suitable for daily intake of

enhanced carotenoid levels are shortenings, cooking fats, butter, margarine and low fat margarine.

In such products, suitably 15-35 mg/kg, preferably 15-35 mg/kg carotenoids selected from the group consisting of  
5 lutein, lycopene, alpha and beta carotene, or mixtures thereof, is present. In a specific embodiment, this type of product contains 5-15 mg/kg of lutein, 1-8 mg/kg of lycopene, present as separate encapsulates or as encapsulates containing at least both, and 3-8 mg/kg of a  
10 mixture of alpha and beta carotene in free form.

In the current invention, the edible compositions, in particular the edible emulsions are not colored strong yellow, pink, orange or red or a mixed-colour thereof  
15 despite the fact that they preferably comprise at least 15 mg/kg carotenoids, which would under circumstances disclosed in the prior art, wherein carotenoids are present in free form without any further treatment lead to  
intensely red, orange or yellow products.

20 It will be appreciated that the Yellowness Index depends on the product concerned.

If spreadable food products like margarine, cheese or products like yoghurt are desired, said products preferably  
25 show a pale yellowish colour, evidenced by a Yellowness Index of from 40 to 70 preferably from 45 to 65.

If products like off-white dressings or mayonnaise are desired, said products are preferably characterised by a Yellowness Index of from 10 to 40.

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The current invention is in particular very suitable for food products containing fat or a fat replacer such as table spreads or margarine or butter or margarine- or butter-like products.

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The terms fats and oils are used interchangeably in this document. Both by fat and oil is meant a triglyceride composition or a non-toxic material having properties comparable with those of triglycerides, which material may be indigestible.

Fat can be of any source such as dairy fat, vegetable fat, fish oil or a combination thereof. Preferred fats have a high content of polyunsaturated fatty acid residues as these fats are considered to have a beneficial effect on cholesterol levels in serum, in addition to the effect provided by sterols.

Fat can for example be selected from the group comprising sunflower oil, safflower oil, palm oil, palm kernel oil, illippe oil, linseed oil, rapeseed oil, linola oil, soy bean oil, coconut oil or combinations thereof.

If a combination of fats is used, it is preferred that the resulting fat blend comprises at least 30 wt% on total fat blend, more preferred at least 45 wt% of polyunsaturated fatty acids.

Preferred products comprise at least 5 wt% fat, more preferred from 5 to 80 wt% fat which is a common amount for products such as margarine.



19

According to a preferred embodiment the edible composition according to the invention comprises from 0.1 to 25 wt% protein, preferably from 2 to 15 wt%, more preferred from 2 to 5 wt% protein.

5

In a recommendable embodiment of the invention, the food products within the scope of the invention are storage stable. This implies that among others, products according to the invention preferably do not show increased colouring of the food products in the course of storage as a result of breaking of encapsulates.

For products according to the invention, the desired storage time depends on regular use. Such a food product need to be storage stable for a time sufficient for transportation to for example a super market, and in addition thereto for a certain time after the product has been bought by a consumer.

Preferred products are storage stable for at least 4 weeks, preferably at least 10 weeks, more preferred around 4 to 9 months. This storage stability implies that preferred products show, throughout the product, when compared to a product obtained directly after production, an increase in Yellowness Index, of less than 10% after 4 weeks, preferably at most 20% after 10 weeks, whereby the product obtained directly after production is the reference product.

In addition to the above-mentioned ingredients, food products according to the invention may optionally contain

further ingredients suitable for use in these products. Examples of these materials are sugar or other sweetener materials, EDTA, spices, salt, bulking agents, egg yolk, emulsifiers, stabilising agents, flavouring materials, colouring materials, acids, preserving agents, vegetable particles etc.

Food products according to the invention may also contain protein. Such protein can for example serve to improve structure or flavour characteristics. Said protein can be derived from any source, for example dairy protein.

Examples of suitable methods to produce fat continuous products like high fat margarine or water continuous products such as cheese are described at a later stage.

The edible compositions comprising encapsulated colored compounds like carotenoids and/or derivatives thereof, can be added to a food product or to ingredients suitable to prepare a food product in any known way, or can be a food product in itself.

In order to obtain a food product of optimal quality, it is advisable that the carotenoid comprising edible composition is homogeneously mixed with edible material or at least with one ingredient of the edible material that forms a food product. A homogeneous distribution is believed to avoid that certain parts of the food products contain high levels of carotenoid, whereas other parts of the food products contain low levels of carotenoids. This may result in a very variable daily intake of these healthy compounds.

21

Said colored compound comprising edible composition can for example be added to the aqueous phase before mixing with the fatty phase (if present) or can be added to the final product under stirring to ensure homogeneous distribution of the carotenoid composition in the final product.

The edible composition can be included in a food product whereby said edible composition is for example in the form of an emulsion or in the form of a powder.

Especially if a process is used wherein high shear regimes are applied, the colored compound comprising edible composition is preferably added just before packaging.

The amount of encapsulated colored compound comprising edible composition that is added to the final food product is such that the amount of colored compound on total weight is at least 15 mg/kg on total product. It is believed to be within the capabilities of the skilled person to calculate the amount of encapsulated composition to be added, starting from the initial amount of colored compound like carotenoid that is present in said encapsulated composition.

According to another embodiment the obtained colored compound comprising edible composition is used for the preparation of a fat continuous product such as a margarine or margarine like products for example comprising from 5 to 80 wt% fat. A preferred process to prepare such a margarine (like) product comprises the steps of emulsification of aqueous phase in a melted fatty phase, mixing the formed emulsion to ensure uniformity, cooling said emulsion in a

shear unit, for example a tubular swept surface heat exchanger, to obtain crystallisation, working the resulting partially crystallised emulsion in for example a pin stirrer unit and packaging the resulting fat continuous product. The encapsulated colored composition is preferably added just before packaging. Optionally before packaging the emulsion is subjected to a resting treatment to increase the final product consistency. Said resting is for example carried out in a resting unit or a quiescent tube. Optionally the aqueous phase is pasteurised before mixing it with a fatty phase.

In the preparation of water free products like shortenings, the encapsulated colored compound comprising edible composition is preferably added in dried, powdered form.

A preferred process to prepare a pumpable oleaginous food product comprises the steps of melting triglyceride fat in a shear mixer such as an A unit, cooling to below the alpha crystallisation temperature and subsequent, or prior to cooling, mixing the triglyceride fat with the above indicated aqueous phase. The resulting product is preferably stored at a temperature from 0 to 25 °C, and preferably between 0 and 15 °C.

Water continuous cheese products like fresh cheese are for example obtained by a process comprising the steps of preparing a cream composition comprising an oil in water emulsion, subjecting this composition to acidification, concentrating the composition under whey removal, applying

23

a homogenising treatment to the obtained concentrate to obtain the final product.

5 A water continuous spreadable product can for example be prepared by the method indicated above for fresh cheese, whereby optionally the whey removal and concentration step are left out.

10 The edible composition comprising encapsulated colored compounds like carotenoids can be added at any stage during processing.

15 A bicontinuous product can suitably be prepared by a process comprising the steps of forming an oil and water containing mixture, homogenising the oil and water mixture under such conditions that an oil in water dispersion is obtained with fat droplets having a preferred volume weighted mean diameter of less than 5 micrometer, cooling the dispersion under conditions such that coalescence of the oil droplets is induced. Cooling can be effected by 20 passing the emulsion through for instance a cooling coil, or a scraped surface heat exchanger.

The encapsulated colored compound comprising edible composition can be added at any stage during processing, preferably after homogenisation.

25

Dressings comprising for example from 0 to 60 wt% fat can suitably be prepared by preparation of an aqueous phase comprising for example flavour components, preservatives, thickeners and emulsifiers. A fatty phase comprising a fat 30 such as sunflower oil, or a fat replacer, enriched with sterol compositions can be added to the aqueous phase under

24

stirring. The obtained mixture is preferably thoroughly mixed to obtain a pre-emulsion and then processed in a low shear device such as a colloid mill. A water continuous gel dispersion then results. The edible composition comprising 5 encapsulated colored compounds is preferably added after the colloid mill treatment.

The invention is now illustrated by the following examples.

25

**Examples****Example A**

*spreads containing 70 wt% of fat were prepared as follows:*

- 5 In this example, amounts indicated are based on the final product.

A fat phase was prepared of ingredients:

- 63% sunflower oil  
10 7% of an interesterified fat composition of 65 parts of a  
mild fractionated palm oil stearine and 35 parts of  
palm kernel oil  
0.04% lecithin  
0.08% emulsifier Hymono 8903<sup>™</sup>  
15 carotene (amount as indicated per example)  
TiO<sub>2</sub> (amount as indicated per example)

Ingredients of the aqueous phase

- 1.75% NaCl  
20 1% sweet whey  
0.04% lactic acid  
0.1% K-sorbate  
water (balance amount)  
pH was set around 5.0

25

The fat phase and aqueous phase were mixed to a pre-mix,  
and passed through an A-A-C processing line under the  
conditions as follows

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The premix was heated to about 60 °C, and passed through the processing line for which the following conditions were applied:

A unit: 800 rpm, at 20 °C,

5 A unit: 800 rpm, at 8 °C

C unit: 200 rpm

The throughput was 4.5 kg/hr

*Preparation of carotenoid encapsulates of protein*

10 Carotenoid encapsulates were prepared as follows:

A 2.5% Na-caseinate solution was prepared by adding 37.5 g of Na-caseinate, 1461 g of demineralized water, and 1.5 g of potassium sorbate, and stirring for one hour.

15

To 1440 g of the so obtained solution, 160 g of a mixture of sunflower oil and  $\beta$ -carotene (4.8 g  $\beta$ -carotene, 155.2 g sunflower oil) was added and mixed, so that a turbid, dark red colored mixture was obtained. The mixture was

20 subsequently treated first by an Ultra Turrax<sup>™</sup> and then by a high pressure homogenisator (pressure of 150 bar) to form an emulsion.

To the emulsion 0.75% D-gluconic acid lactone was added to lower the pH. The mixture was stirred in a vessel at 400 rpm for 45 minutes, and at 1800 rpm for another 45 minutes.

To the obtained mixture, 7.83 transglutaminase was added (1% enzyme), the mixture was stirred at 400 rpm for 17.5 hours.

30 Subsequent centrifuging, filtering, washing, and repeating this, resulted in encapsulates containing 3010 mg/kg  $\beta$ -



27

carotene. The encapsulates obtained are referred to as type "A".

*Preparation of carotenoid encapsulates of protein*  
5 containing  $TiO_2$ .

The procedure of the preparation of carotenoid encapsulates of protein is repeated, the only alternative to the procedure being that after addition and mixing of the sunflower oil and carotenoids to the solution, 36 grams of  
10  $TiO_2$  were added. The Ultra Turrax and homogenisator treatment were carried out as described above. As a result, encapsulates containing 8%  $TiO_2$  and 3214 mg/kg of  $\beta$ -carotene were obtained.

The encapsulates obtained are referred to as encapsulates  
15 type "B".

*Preparation of complex coacervates*

Carotenoid encapsulates were prepared as follows:

20 600 grams of a gelatin solution (concentration 2%, temperature about 60 °C) were added to 60 grams of a sunflower oil containing  $\beta$ -carotene (at 60 °C), and the mixture was stirred in an Ultra Turrax™. The amount of  $\beta$ -carotene was set so as to obtain a concentration of 1% in  
25 the sunflower oil. To the mixture, 600 grams of a 2 wt% aqueous solution of Arabic Gum were added, the temperature was kept at 60 °C. After the addition of the Arabic Gum solution, an 28 grams of  $TiO_2$  were added under stirring. The pH was set at 4.3, using HCl (4 N), while the stirring  
30 was continued. The obtained mixture was allowed to cool to

20 °C under slow stirring, and subsequently cooled to about 4 °C.

The obtained encapsulates (coacervates) containing composition was filtered, and the encapsulates were  
5 obtained. To the encapsulates, water was added, the temperature of the composition was raised to 50 °C, filtered again and the encapsulates where then washed with water.

To the coacervates, about 500 ml of water was added. Under  
10 slow stirring, 4.5 ml glutaraldehyde was added. Stirring was continued for about 19 hours.

By filtration and washing, 151 g of encapsulates was obtained. The  $\beta$ -carotene was present in an amount of about 1% of the total weight of the particles.  $\text{TiO}_2$  was present  
15 in an amount of about 30% based on total particle weight. The encapsulates obtained are referred to as encapsulates type "C".

*Preparation of complex coacervates containing  $\text{TiO}_2$*

20 The procedure of example IC was repeated, with the proviso that the amount of  $\text{TiO}_2$  was 9.33 grams.  
The encapsulates obtained are referred to as encapsulates type "D".

**Comparative Examples I-IV**

Spreads were prepared following the procedure as described above. Carotene was present in the in free form, i.e. not encapsulated, in amounts as indicated. No  $\text{TiO}_2$  was used.

- 5 The carotenoids level on total spread was measured after the spread was prepared, and is indicated in Table I.

**TABLE I**

Ex.	$\beta$ -carotene (mg/kg)	$\text{TiO}_2$	Yellowness Index	Yellowness Factor (g/kg)
I	22.4	none	92	4098
II	56.5	none	111	1965
III	104.7	none	125	1194
IV	153.5	none	134	873

10 **Example V-VII**

Spreads were prepared as described in the above procedure. This time, to the fat phase of the spreads, encapsulates of type "A" were added in an amount as indicated in Table II. No  $\text{TiO}_2$  was used.

15

**TABLE II**

Ex.	$\beta$ -carotene (mg/kg)	$\text{TiO}_2$	Yellowness Index	Yellowness Factor (g/kg)
V	17.6	none	59	3352
VI	35.1	none	68	1937
VII	103.1	none	88	853

It was observed that with these particles, it is preferred to add these after having passed the spread through the Votator processing line, under gentle stirring.

30

**Example VIII-IX**

Spreads were prepared as described in the above procedure. This time, to the fat phase of the spreads, encapsulates of type "B" were added in an amount resulting in spreads

5 containing an amount of the carotene as indicated in Table III.

**TABLE III**

Ex.	$\beta$ -carotene (mg/kg)	TiO <sub>2</sub> present	Yellowness Index	Yellowness Factor (g/kg)
VIII	25.4	yes	70	2756
IX	62.8	yes	90	1433

10 **Example X-XII**

Spreads were prepared as described in the above procedure. This time, to the fat phase of the spreads, encapsulates of type "C" were added in an amount in an amount resulting in spreads containing an amount of the carotene as indicated

15 in Table IV.

**TABLE IV**

Ex.	$\beta$ -carotene (mg/kg)	TiO <sub>2</sub> present	Yellowness Index	Yellowness Factor (g/kg)
X	37.4	yes	69.5	1858
XI	66	yes	83	1257
XII	48	yes	87	1813

20

31

**Example XIII-XVI**

Spreads were prepared as described in the above procedure. This time, to the fat phase of the spreads, encapsulates of type "D" were added in an amount resulting in spreads containing an amount of the carotene as indicated in Table V.

**TABLE V**

Ex.	$\beta$ -carotene (mg/kg)	TiO <sub>2</sub>	Yellowness Index	Yellowness Factor (g/kg)
XIII	15.0	Yes	31.34	2089
XIV	44.3	Yes	43.96	998
XV	95.1	Yes	50.13	527
XVI	158	yes	60.61	384

10

**Example B**

Spreads containing 40 wt% of fat were prepared as follows:

15 A fat blend was prepared of ingredients, amounts based on amount of fat blend:

73% bean oil

17% of an interesterified fat composition of hardened palmkernel oil

20 10% palm oil

This fat blend was used for the preparation of a fat phase as follows (amounts are based on total end product).

39.78 % of the above fat blend

25 0.05% lecithin

32

0.16 emulsifier (diglyceride of hardened palmoil)

0.012 carotene

Ingredients of the aqueous phase

5 1.1% gelatin

0.48% NaCl

0.27% acidic whey

0.12% K-sorbate

water (balance amount)

10 pH was set around 5.0 with citric acid

The fat phase and aqueous phase were mixed to a pre-mix, and passed through an A-A-A-C processing line under the conditions as follows

15 The premix was heated to about 60 °C, and passed through the processing line for which the following conditions were applied:

A unit: 1000 rpm, at 20 °C,

A unit: 1000 rpm, at 14 °C

20 A unit: 1000 rpm, at 9 °C

C unit: 900 rpm

The throughput was 150 kg/hr

Nitrogen was inserted, after which gentle mixing took  
25 place. Amount of Nitrogen is as indicated in the example.

The amount of carotene was measured, and the yellowness index was measured as indicated in the patent specification. The yellowness factor was determined by  
30 deviding the carotene level with the outcome of the

yellowness index measurement. The results are found in table VI.

5

TABLE VI

Ex.	$\beta$ -carotene (mg/kg)	N <sub>2</sub> Present %	Yellowness Index	Yellowness Factor (g/kg)
XVII*	18	0	77.3	4294
XVIII	18	17	68	3778
XIX	18	23	60	3333
XX	18	41	56	3111

\* Comparative example

This example shows that nitrogen is suitable for reducing colour of free carotenoids present in the food emulsion, at  
10 nitrogen levels significantly higher than applied in the past.

Claims

1. Edible composition containing at least 15 mg/kg  
carotenoids, and preferably at least 18 mg/kg, and  
5 having a yellowness factor as defined in this  
specification of less than 4000, and have a yellowness  
index in the range of 1 to 90, the carotenoids being  
evenly distributed throughout the composition.
- 10 2. Edible composition according to claim 1, wherein the  
composition has yellowness index in the range of 1 to  
75.
- 15 3. Edible composition according to claim 2, wherein the  
composition has a yellowness factor of less than 3333,  
preferably of less than 2850, and a yellowness index in  
the range of 1 to 65.
- 20 4. Edible composition according to claim 3, wherein at  
least 20 mg/kg carotenoids are present, and wherein the  
composition has a yellowness factor of less than 2850,  
and a yellowness index in the range of 1-70.
- 25 5. Edible composition according to claim 1, wherein at  
least 45 mg/kg carotenoids are present, and wherein the  
composition has a yellowness factor of less than 2000,  
and a yellowness index in the range of 1-90.
- 30 6. Edible composition according to any of the preceding  
claims, wherein the composition is an edible emulsion.



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7. Edible composition according to any of the preceding claims, wherein the carotenoids are present in encapsulated form.
- 5 8. Edible composition according to claim 7, wherein the carotenoids encapsulates are complex coacervates.
9. Edible composition according to claim 8, wherein the complex coacervates contain gelatin and Arabic gum.
- 10 10. Edible composition according to claim 7, wherein the encapsulates have a coating of at least one protein layer.
- 15 11. Edible composition according to claim 10, wherein the encapsulates have at least 3 protein layers.
12. Edible composition according to any of claims 7 to 11, wherein the encapsulate contains at least 5-30 wt%  $\text{TiO}_2$ , based on the weight of the total encapsulate particle.
- 20 13. Edible composition according to claim 12, wherein the encapsulate contains 5-20 wt%  $\text{TiO}_2$ , based on the weight of the total encapsulate particle.
- 25 14. Edible composition to any of the preceding claims, wherein the edible composition is an edible emulsion.
- 30 15. Fat based food product containing an edible composition according to any of claims 1-14.

16. Fat based food product according to claim 15, wherein  
the food product is selected from the group of  
shortenings, cooking fats, cheese, dressings, including  
mayonnaise, ice cream, milk type drinks, (drink)  
5 yoghurt, toppings and fillings, butter, margarine and  
low fat margarine.
17. Fat based food product according to claim 16, wherein  
the food product is selected from the group of  
10 shortenings, cooking fats, butter, margarine and low  
fat margarine.
18. Fat based food product according to claim 17, wherein  
the food product contains 15-25 mg/kg carotenoids  
15 selected from the group consisting of lutein, lycopene,  
alpha and beta carotene, or mixtures thereof.
19. Fat based food product according to claim 18 wherein  
the food product contains 5-15 mg/kg of lutein , 1-8  
20 mg/kg of lycopene, present as separate encapsulates or  
as encapsulates containing both, and 3-8 mg/kg of a  
mixture of alpha and beta carotene in free form.
20. Encapsulates containing carotenoids, characterised in  
25 that the carotenoids are encapsulated in complex  
coacervates of at least two polymers, and the  
encapsulates containing 5-30 wt% on particle weight, of  
titaniumdioxide.

37

**Abstract**

The invention is about an edible composition, preferably an edible emulsion, which contains high amounts of  
5 carotenoids, but is not colored in a manner as expected when using such high amounts of carotenoids, as defined by the composition having a yellowness factor of less than 4000 g/kg, and a yellowness index in the range of 1 to 90.

